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Risk Factors for Acute Non-A, Non-B Hepatitis and Their Relationship to Antibodies for Hepatitis C Virus: A Case–Control Study

Alfonso Mele, MD, Luciano Sagliocca, MD, Giuseppe Manzillo, MD, Francesco Converti, MD, Pietro Amoroso, MD, Maria Antonietta Stazi, MSc, Luigina Ferrigno, Mariella Rapicetta, MD, Elisabetta Franco, MD, Brunella Adamo, MD, Filippo Palumbo, MD, Costanza Sbreglia, MD, Augusto Panà, MD, Paolo Pasquini, MD, and the SEIEVA Collaborating Group

ABSTRACT

A case–control study was carried out comparing 333 case subjects with non-A, non-B hepatitis and 1095 hospital control subjects. Of 333 case subjects, 197 (59%) were positive for hepatitis C antibody (anti-HCV). Excluding blood transfusion and intravenous drug use, surgical intervention and dental therapy were strongly associated with anti-HCV–positive cases: in particular, obstetric and gynecology surgical intervention was found to be strongly associated with HCV positivity (odds ratio [OR] = 32; 95% confidence interval [CI] = 7, 147). Raw shellfish consumption was a risk factor for anti-HCV–negative cases (OR = 2.2; 95% CI = 1.0, 5.1), thus suggesting an enterically transmitted virus in sporadic non-A, non-B hepatitis in Italy. (*Am J Public Health.* 1994;84:1640–1643)

Introduction

Non-A, non-B hepatitis includes several agents that are both blood and enterically transmitted.[1–3] A large number of patients with blood-transmitted non-A, non-B hepatitis may develop chronic liver disease. Blood transfusions and intravenous drug use explain fewer than 50% of the hepatitis cases; therefore, identification of additional risk factors for the disease is necessary.[4,5]

Risk factors associated with non-A, non-B hepatitis were evaluated in a case–control study based in three Naples hospitals (Italy) from November 1987 through May 1991.

Study Population and Methods

All case subjects were recruited in an infectious-diseases hospital, had serum

Alfonso Mele, Maria Antonietta Stazi, Luigina Ferrigno, and Paolo Pasquini are with the Istituto Superiore di Sanità, Laboratorio di Epidemiologia e Biostatistica, Roma. Luciano Sagliocca, Brunella Adamo, and Filippo Palumbo are with the Regione Campania, Osservatorio Epidemiologico, Napoli. Giuseppe Manzillo, Pietro Amoroso, and Costanza Sbreglia are with the Ospedale Cotugno, Napoli. Francesco Converti is with the Regione Campania, Servizio Epidemiologico USL 40, Napoli. Mariella Rapicetta is with the Istituto Superiore di Sanità, Laboratorio di Virologia, Roma. Elisabetta Franco is with the Cattedra di Igiene, Università della Basilicata, Potenza. Augusto Panà is with the Cattedra di Igiene, Università "Tor Vergata," Roma. For a list of the SEIEVA Collaborating Group, see the Acknowledgments.

Requests for reprints should be sent to Alfonso Mele, MD, Istituto Superiore di Sanità, Laboratorio di Epidemiologia e Biostatistica, Reparto di Epidemiologia Clinica, Viale Regina Elena 299, 00161 Roma Italy.

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aminotransferase levels at least three times greater than the upper limit of normal values, and were negative for immunoglobulin M (IgM) antibodies to hepatitis A virus and hepatitis B core antigen (radioimmunoassay method, Abbott Laboratories). No case subject had a history or clinical sign of hepatic injury due to medication, alcohol, or other hepatobiliar diseases, and case subjects had an onset of disease not longer than 2 weeks.

Non-A, non-B case subjects with IgM antibodies against cytomegalovirus (Abbott CMV-M enzyme immunoassay) or with IgM antibodies against Epstein–Barr virus (Ortho monolert) were excluded from the study.

Serum samples from all case subjects were stored at −80°C.

Assays for hepatitis C antibodies (anti-HCV) were done before 1990 with the first-generation enzyme-linked immunosorbent assay (ELISA) (Ortho Diagnostics). Thereafter, the second-generation test was applied to all new cases and to serum samples that were negative for anti-HCV by the first-generation ELISA. Serum samples that showed an optical density between 0.7 and 1.7 were retested by supplemental second-generation recombinant immunoblot assay (RIBA) and were considered positive when two or more bands were present.

All anti-HCV–negative case subjects recruited from 1990 were followed-up for at least 6 months (range, 6–15 months).

The following control subjects were chosen: (1) patients without bloodborne and enterically transmitted disease admitted to the same hospital as the case subjects; (2) patients from the neurosurgery and ear, nose, and throat wards of a pediatric hospital; and (3) patients from the emergency, orthopedics, and surgery wards of a general hospital. Both the pediatric and general hospitals were located in the same area as the infectious-diseases hospital.

Case and control subjects lived in the same area and were interviewed on admission to the hospitals. Data for children were obtained from the mothers.

Exposure within 6 months and 6 weeks of the onset of disease was investigated for parenteral risk factors (including sexual exposure) and raw shellfish consumption, respectively, by means of a standardized questionnaire.

Age groups used for analysis were 0–9, 10–14, 15–19, 20–34, 35–54, and ≥ 55 years.

TABLE 1—General Characteristics of Control and Case Subjects, Naples, Italy, 1987 through 1991

	Subjects, %		
		Case	
Age, y	Control (n = 1095)	Positive for Hepatitis C Virus Antibody (n = 197)	Negative for Hepatitis C Virus Antibody (n = 136)
0–9	18.9	1.0	13.2
10–14	8.8	0.5	12.5
15–19	13.6	14.2	19.1
20–34	20.2	64.5	39.0
35–54	20.8	11.7	14.0
55 and over	17.7	8.1	2.2

TABLE 2—Diagnoses of Control Subjects (n = 1095)

Diagnosis	Percentage
Infectious diseases (Measles, mumps, brucellosis, rickettzial diseases, respiratory tract infections, encephalitis, herpes zoster, varicella, mononucleosis, toxoplasmosis)	13.3
Diseases following trauma and emergency surgery (Fractures, dislocations, abdominal trauma, acute appendicitis, cranial trauma)	22.9
Diseases undergoing surgical treatment (Orthopedic surgery, anorectal diseases, thyroid diseases, breast benign diseases, inguinal hernia)	32.1
Ear, nose, and throat diseases	9.7
Others (Inflammatory gastrointestinal diseases, depression, or arterial peripheral diseases, respiratory diseases, renal colic, irritable bowel syndrome)	21.9

Note. Two hundred twenty-one patients were recruited in the infectious-diseases hospital.

The polychotomous logistic technique was used to estimate the independent effect of the studied exposure variables on the risk of being either anti-HCV positive or anti-HCV negative.[6,7]

We first considered intravenous drug exposure, blood transfusion before and after 1990, age, sex, and education and thereafter the effects of all other exposure variables.

First-order interaction terms were always tested.

Results

Three hundred and forty-two case subjects and 1095 control subjects were recruited. Of the 342 case subjects, 251 (73%) were jaundiced; serum samples were taken from all case subjects within 21 days of the onset of disease. Four case subjects positive for IgM antibodies against cytomegalovirus and five positive for IgM antibodies against Epstein–Barr virus were excluded.

Of 333 case subjects, 187 (56%) were anti-HCV positive by the first- or second-generation test at first blood drawn (70 of 148 negative samples at first-generation ELISA were positive at second-generation ELISA and two of eight serum samples with an optical density between 0.7 and 1.7 were confirmed by second-generation RIBA).

Among the 68 anti-HCV–negative subjects followed-up, 10 (15%) became anti-HCV positive (second-generation test, second-generation RIBA confirmed): 5 subjects after 45 days from the onset of disease, 3 subjects after 2 months, and 2 subjects after 4 months. Overall, 164 of 248 (66%) males and 33 of 85 (39%) females were anti-HCV positive.

TABLE 3—Case and Control Subjects Exposed to Intravenous Drug Use and Blood Transfusion, Naples, Italy, 1987 through 1991

	Control Subjects		Case Subjects					
			Anti-HCV Positive			Anti-HCV Negative		
Risk Factor	%	Total No.[a]	%	Total No.[a]	OR (95% CI)	%	Total No.[a]	OR (95% CI)
Intravenous drug use	0.7	1086	51.3	195	71.0 (32.0, 160.0)	13.3	135	13.0 (5.3, 31.0)
Blood transfusion (1987–1989)	0.8	759	14.8	128	49.0 (17.0, 140.0)	4.6	87	7.0 (1.9, 27.0)
Blood transfusion (1990–1991)	0.3	333	1.4	69	1.9 (0.01, 270.0)	0.0	48	Not calculable

Note. Adjusted ORs (95% CIs) were estimated by polychotomous logistic regression according to positivity for hepatitis C virus antibody (anti-HCV). ORs were adjusted for age, sex, education level, and the other listed variables.
[a]Discrepancies in the numbers are due to missing values ("I don't know" answer).

TABLE 4—Case and Control Subjects Exposed to Considered Risk Factors, after Subjects with Blood Transfusions and Intravenous Drug Users are Excluded, Naples, Italy, 1987 through 1991

	Control Subjects		Case Subjects					
			Anti-HCV Positive			Anti-HCV Negative		
Risk Factor	%	Total No.[a]	%	Total No.[a]	OR (95% CI)	%	Total No.[a]	OR (95% CI)
Surgical intervention	2.4	1077	16.5	79	12.0 (4.0, 35.0)	12.4	113	4.9 (2.0, 12.0)
Hospitalization	10.3	1079	22.8	79	0.8 (0.3, 2.0)	17.5	114	1.1 (0.6, 2.3)
Dental therapy	11.0	1079	22.4	76	1.9 (1.0, 3.5)	13.2	114	1.2 (0.6, 2.1)
Other percutaneous exposures[b]	2.1	1072	5.1	78	1.7 (0.5, 6.0)	5.4	111	1.8 (0.7, 4.8)
More than one sexual partner[c]	6.6	775	16.9	77	1.4 (0.7, 3.0)	11.5	78	1.2 (0.5, 2.7)
Raw shellfish	5.7	715	16.7	54	1.4 (0.6, 3.6)	13.6	66	2.2 (1.0, 5.1)

Note. Adjusted ORs (95% CIs) were estimated by polychotomous logistic regression according to positivity for hepatitis C virus antibody (anti-HCV). ORs were adjusted for age, sex, education level, and the other listed variables.
[a]Discrepancies in the numbers are due to missing values ("I don't know" answer).
[b]Ear piercing, tattooing, acupuncture, electrolysis, or attendance at a chiropodist or manicurist.
[c]Age greater than 14 years.

In Tables 1 and 2 general characteristics of case and control subjects are presented: 26% of the anti-HCV–negative case subjects vs 1.5% of the anti-HCV–positive ones were younger than 15 years.

Among the case subjects, 118 were intravenous drug users and 24 received blood transfusions. The odds ratios were much higher for both these risk factors among anti-HCV–positive case subjects than among anti-HCV–negative ones (Table 3).

Of 215 case subjects, 23 (11%) reported receiving blood transfusions from 1987 through 1989; 1 of 117 case subjects (0.9%) received a blood transfusion after 1989.

Two anti-HCV–positive case subjects who were intravenous drug users received blood transfusions, 3 had surgery, and 43 had multiple sexual partners. Of the 20 anti-HCV–positive case subjects who received blood transfusions, 15 also had surgery.

After excluding blood transfusion and drug use, surgical intervention was significantly associated in both case series, and the risk was higher in anti-HCV–positive case subjects than in the anti-HCV–negative ones (Table 4). The strongest association was found in female case subjects age 15–44 years for obstetric and gynecology interventions: odds ratio (OR) = 32.0 (95% confidence interval [CI] = 7.5, 147.0) for anti-HCV–positive cases; OR = 16.5 (95% CI = 4.0, 73.0) for anti-HCV–negative cases. When control subjects undergoing surgical treatment were excluded, the ORs were 18 (95% CI = 5.1, 66.0) among anti-HCV–positive cases and 7.5 (95% CI = 2.6, 22.0) among anti-HCV–negative cases.

Dental therapy was associated with anti-HCV–positive cases, and a nonsignificant association was found between other percutaneous exposures and both cases series. A small but not significant excess of risk (OR = 1.4; 95% CI = 0.7, 3.0) was found among anti-HCV–positive case subjects exposed to multiple heterosexual partners (Table 4).

Thirty-three of 197 (17%) anti-HCV–positive case subjects and 66 of 136 (49%) anti-HCV–negative case subjects did not report any considered parenteral exposure.

A significant association existed between raw shellfish consumption and anti-HCV–negative cases. Raw shellfish consumption was the only risk factor for 7 of 66 (11%) anti-HCV–negative case subjects vs 3 of 54 (6%) anti-HCV–positive case subjects (Table 4).

No effect modification was found between the considered risk factors.

Discussion

More than 50% of the case subjects with non-A, non-B hepatitis were anti-HCV positive, but the diagnosis of acute hepatitis C is reliable only for the 10 case

subjects of 68 (15%) who seroconverted during the follow-up.

Blood transfusion and intravenous drug use account for 61% of the anti-HCV–positive cases and 16% of the anti-HCV–negative cases. Surgical intervention, dental therapy, and other percutaneous exposures are additional parenteral risk factors found to be associated with non-A, non-B hepatitis. Interestingly, the strongest association was shown with obstetric and gynecological interventions. Given that a large proportion of the general population is exposed to these risk factors, the present results and those reported for hepatitis B[8] underscore the importance of implementing efficient procedures for sterilization of instruments and the use of disposable materials, especially when a high turnover of patients combined with emergency surgical intervention (such as in gynecology) represents an increased risk of infection.

Few case subjects with non-A, non-B hepatitis and a history of blood transfusion were enrolled after 1989, when the anti-HCV test was introduced in the screening of blood banks. That fact suggests that a high number of non-A, non-B cases of hepatitis in patients reporting a history of blood transfusion were due to hepatitis C virus.

Heterosexual transmission of hepatitis B, non-A, non-B, and C viruses has been outlined in previous studies.[9–11] Our data show a slight (OR = 1.4; 95% CI = 0.7, 3.0) but not significant excess of risk for anti-HCV–positive case subjects exposed to multiple sexual partners.

The estimated odds ratios for blood transfusions, intravenous drug use, surgical intervention, and dental therapy were higher in the anti-HCV–positive cases. Although the anti-HCV test applied in this study does not discriminate acute from past infection, anti-HCV positivity elicits a group of non-A, non-B case subjects older than the anti-HCV–negative case subjects, and with greater exposure to parenteral risk factors. At present no test for additional non-A, non-B parenteral viruses is available, but it is likely that the anti-HCV–positive group includes acute non-A, non-B cases due to other blood-transmitted and orofecally transmitted viruses. At the same time, the anti-HCV–negative group might include hepatitis C cases.

The association of raw shellfish consumption with anti-HCV–negative cases suggests the possible presence of enterically transmitted non-A, non-B hepatitis in Italy.

Case subjects did not include all the patients with non-A, non-B hepatitis notified in the area, and hospital control subjects were selected in an attempt to avoid the selection process culminating with cases being diagnosed in the study hospital. The pediatric and general hospitals were located in the same area as the infectious-diseases hospital, where the control subjects would have been admitted had they been hospitalized with acute hepatitis. Furthermore, the broad diagnostic representation of control subjects probably prevented control selection bias.

The inclusion of patients undergoing surgical intervention as control subjects might have led to an underestimation of odds ratios: the associations between cases and surgical intervention were stronger after omitting these control subjects.

Control subjects with infectious diseases such as mononucleosis (n = 5) can share some risk factors with case subjects. When these control subjects were excluded, the estimate of odds ratios remained unchanged. □

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The SEIEVA Collaborating Group consists of select staff of the following institutions: the Regione Campania, Osservatorio Epidemiologico, Napoli; the Ospedale Cotugno, Napoli; the Regione Campania, Servizio Epidemiologico USL 40, Napoli; the Istituto Superiore di Sanità, Laboratorio di Virologia, Roma; and the Cattedra di Igiene, Università "Tor Vergata," Roma.

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